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Numbers of Tufted Ducks *Aythya fuligula* wintering at Lough Neagh declined dramatically following the winter of 2000/01. The abundance and biomass of benthic macroinvertebrates, their main food source, declined significantly between the winters of 1997/98 and 2010. Therefore, information on recent diet was required to determine if there had been any significant changes before and after the observed declines in numbers of both macroinvertebrates and birds. Here, we used oesophageal content analysis to characterise the contemporary diet of Tufted Ducks at Lough Neagh during 2010-12. Out of 75 shot ducks, only three individuals had prey items in their oesophagi while all four ducks that accidentally drowned in gill nets contained prey items. Oesophageal contents were then compared with data collected during a study conducted in the late 1990s. Contemporary diet of Tufted Ducks was dominated by *Asellus aquaticus* (48%), but molluscs (14%), grain (13%) and chironomid larvae (11%) were also consumed. Between 1998-99 and 2010-12, the contribution of *Asellus aquaticus* to the diet significantly decreased while the proportions of chironomid larvae, grain, *Gammarus* spp. and *Mysis* spp. increased. Alternative methods of dietary analysis, for

example stable isotope analysis, are recommended in future studies of diving duck diet at Lough Neagh.

Introduction

Dietary analysis provides information on foraging behaviour, habitat use and general ecology. Temporal variation in diet may contribute to our understanding of population dynamics, particularly in species exhibiting declining populations. For some species, such as diving ducks, diet cannot be inferred from direct observations of foraging, and alternative methods of dietary analysis must be employed; typically stomach content analysis. Analysing stomach contents is simple and straightforward, providing a precise snapshot of recent ingestion (Hynes 1950). However, stomach content analysis tends to be biased towards food items that are hard-bodied and take longer to digest, while softer-bodied prey items which are more easily digested may be under-recorded (Hyslop 1980). This can be overcome to some extent by examining oesophagus contents rather than gizzard contents (Swanson & Bartonek 1970, Guillemette *et al*. 1994). In ducks, prey items pass through the oesophagus undigested, before they enter the proventriculus and gizzard. Ducks swallow and store large amounts of grit in their gizzards (Tománková *et al*. 2013a) to grind up food and aid digestion, causing prey items to breakdown rapidly. Swanson and Bartonek (1970) found that within ten minutes of ingestion, 100% of amphipods, 82% of molluscs and 24% of diptera larvae in the gizzard were digested beyond recognition.

The diet of diving ducks overwintering at Lough Neagh, Northern Ireland was described in a series of detailed studies during the late 20th century (e.g. Winfield & Winfield 1994, Bigsby 2000, Evans 2000). At that time, the diet of Pochard *Aythya ferina*, Scaup *Aythya*

51 *marila* and Goldeneye *Bucephala clangula* was typically dominated by chironomid
52 (Chironomidae) larvae (Winfield & Winfield 1994, Evans 2000) while the diet of Tufted Duck
53 *Aythya fuligula* also contained substantial quantities of molluscs (Winfield & Winfield 1994).
54 Bigsby (2000) suggested that the predominant prey items in the diet of Pochard and Scaup
55 were larval and pupal chironomids, while Tufted Duck fed primarily on *Asellus aquaticus*.
56 Goldeneye took both chironomid larvae and *Asellus aquaticus* in approximately equal
57 proportions (Bigsby 2000).

58 Lough Neagh is a stronghold for overwintering diving ducks in Northern Ireland (Holt
59 *et al*. 2012) and was formerly the most important overwintering site in the United Kingdom
60 for Pochard, Tufted Duck, Scaup and Goldeneye (Pollitt *et al*. 2000). Numbers of migratory
61 diving ducks overwintering at Lough Neagh have declined dramatically since the winter of
62 2000/01. Declines occurred over a period of two winters, after which populations appear to
63 stabilise at relatively low abundances, suggesting the change was rapid (Tománková *et al*.
64 2013b). Between the winters of 1994/95-1998/99 and 2006/07-2010/11, the winter five-year
65 mean declined from 24,525 to 7,599 individuals in Pochard, from 22,566 to 6,938 individuals
66 in Tufted Duck and from 7,557 to 3,501 individuals in Goldeneye (Pollitt *et al*. 2000, Holt *et al*.
67 2012). The density and biomass of macroinvertebrates in Lough Neagh declined by 65-70%
68 between the winters of 1997/98 (prior to observed declines in diving duck numbers) and 2010
69 (after the decline) Tománková *et al*. 2014). These declines may have reduced the quantity and
70 quality of food resources available to overwintering ducks.

71 This study examined the diet of Tufted Ducks overwintering at Lough Neagh using
72 oesophageal contents analysis. The results were compared with similar data gathered during
73 earlier periods of 1988-1990 (Winfield & Winfield 1994), 1998-1999 (Evans 2000) and 1998-

2000 (Bigsby 2000). This comparison was required to determine if there has been any significant temporal change in the composition of the diet of diving ducks.

Methods

Diving ducks (Pochard, Tufted Duck, Scaup and Goldeneye) were obtained from wildfowlers or fishermen operating on Lough Neagh between the winters 2008/09 and 2011/12. Ducks were aged and sexed based on plumage characteristics (Boyd *et al*. 1975, Baker 1993). A total of 79 individuals were collected, of which 75 (95%) had been shot and four (5%) accidentally caught in legally set gill nets. Only seven (9%) ducks examined had prey in their oesophagi. They included all four ducks that had been recovered drowned from gill nets, and only three (4%) that had been shot. All seven individuals were male Tufted Duck; three were first-winter birds and four were adults.

The oesophagus of each duck was dissected and any prey items removed. These were stored in 80% ethanol and subsequently identified. Chironomid larvae head capsules were mounted with either Euparal or Hydromatrix solutions (Brooks *et al*. 2007) to allow identification (Wiederholm 1983, Brooks *et al*. 2007). Results were compared with those from an earlier study on the diet of diving ducks on Lough Neagh investigated in 1998-99 (Bigsby 2000).

Chironomid larvae dissected from the oesophagi of ducks were measured and length frequencies compared with those of chironomids found in oesophagi of Tufted Ducks collected in 1988-1990 (Winfield & Winfield 1994), 1998-1999 (Evans 2000) and 1998-2000 (Bigsby 2000). Data from these three studies were extracted from graphs using Plot Digitizer (version 2.5.1) software. Chironomid larvae were pooled irrespective of species or genus.

To examine changes in the proportional composition of macroinvertebrates in the diet of Tufted Ducks between 1998-99 and 2010-12, 2x2 χ^2 contingency tests were used within each prey category. To determine whether there was any difference in the size of chironomids consumed, their length frequencies were compared between 1988-1990, 1998-99, 1998-2000 and 2010-2012 using a Generalized Linear Mixed Model (GLMM) with a fitted gamma distribution and logarithmic link function where chironomid size was fitted as the dependent variable, study as a fixed factor and size category as a random factor to account for the multiple observations per size category, thus avoiding pseudo-replication.

Results

The diet of Tufted Ducks (Figure 1) was predominantly composed of the freshwater crustacean *Asellus aquaticus* (48%), molluscs (14%) and grain (13%). Of the chironomid larvae, the most commonly consumed genera were *Glyptotendipes* spp. (5%) and *Microtendipes* spp. (4%). Molluscs taken were (in descending order of abundance): *Potamopyrgus antipodarum*, *Lymnaea peregra*, *Valvata* spp., *Planorbis carinatus*, *Physa* spp., *Planorbis corneus* and *Bithynia* spp. Trichoptera larvae belonged to families Leptoceridae, Molannidae, Phryganeidae and Polycentropidae while Hemiptera were of the family Corixidae.

Acknowledging that the sample size was low, the oesophageal contents of Tufted Ducks in 2010-12 differed significantly from 1998-99. Specifically, the greatest change was the appearance of grain in the diet (13%) during 2010-12, which mostly comprised wheat *Triticum* spp., and which was entirely absent in 1998-99 (χ^2_1 = 13.9, P = <0.001). The proportion of chironomid larvae (all genera pooled) increased from 3% to 11% in 2010-12 (χ^2_1 = 4.92, P = 0.027), *Mysis* spp. increased from 2% to 9% (χ^2_1 = 4.71, P = 0.030) and *Gammarus*

spp. increased from 0% to 4% ($\chi^2_1 = 4.08$, P = 0.043) (Figure 1). Conversely, the proportion of *Asellus aquaticus* decreased from 73% during 1998-99 to 48% during 2010-12 ($\chi^2_1 = 13.1$, P = <0.001) (Figure 1). The proportion of molluscs present in the diet did not differ significantly with a proportion of 13% during 1998-99 and 14% during 2010-12 ($\chi^2_1 = 0.043$, P = 0.836) nor did the remaining dietary items being 8% in 1998-99 and 2% in 2010-12 ($\chi^2_1 = 3.79$, P = 0.052) (Figure 1). There was a strong trend for the size frequency distribution of chironomid prey to vary between studies conducted during 1988-90, 1998-99, 1998-00 and 2010-12 ($F_{3,47} = 2.314$, P = 0.088). Specifically, the median length of chironomid larvae in 1998-99 and 2010-12 was smaller than, and exhibited a normal distribution, those during 1988-90 and 1998-00, which exhibited a bimodal distribution (Figure 2).

Discussion

In the current study, all ducks that had been drowned contained prey in their oesophagi compared to only 4% that had been shot. Diving ducks at Lough Neagh, with the exception of Goldeneye, are primarily nocturnal feeders (Evans & Day 2001), so most birds would have finished feeding several hours before being shot by wildfowlers during daylight hours. Consequently, oesophageal contents analysis of shot ducks cannot be recommended in dietary studies. Ducks accidentally caught in gill nets that are captured while feeding would be the best source of material for oesophageal analysis (Tománková *et al.* 2013a); though they are more difficult and less reliable to obtain. Such bias in sampling methods has been reported previously by Winfield & Winfield (1994) and Bigsby (2000).

Analysis of the oesophageal contents of male Tufted Ducks suggested they fed primarily on *Asellus aquaticus* and to a lesser degree on molluscs. Whilst this analysis was

146 based on a small sample size (n = 7), Bigsby (2000) reported a similar diet utilizing a larger,
147 but still limited, sample (n = 18). Evans (2000) and Winfield & Winfield (1994) found that,
148 although not the dominant prey item, *Asellus aquaticus* appeared more important in the diet
149 of Tufted Duck than in other diving duck species overwintering at Lough Neagh. Differences
150 between the studies may be accounted for by changes in food availability which can influence
151 consumer prey choice (Vickery *et al*. 1995). Grain was found in the oesophagi of shot Tufted
152 Ducks in 2010-12 while none was recorded in 1998-99 (Bigsby 2000). It is possible that Tufted
153 Ducks now rely more on supplementary feeding provided by wildfowlers than in the past due
154 to the decline of their macroinvertebrate prey (Tománková *et al*. 2014).

155 The trend for difference in size frequency distribution of chironomid larvae between
156 studies may reflect the timing or location of sampling. The majority of ducks sampled in
157 previous studies (Winfield & Winfield 1994, Bigsby 2000, Evans 2000) had been drowned
158 accidentally in gill nets whilst three out of seven ducks (43%) in the current study had been
159 shot. The differences in the size frequencies of chironomids might be due to the varying depth
160 at which the gill nets were set (which was unknown in most cases) when the birds were
161 caught. Depth may influence the dynamics and thus size structure of chironomid populations
162 (Winfield & Winfield 1994). Chironomid larvae were also pooled irrespective of their
163 taxonomic classification, and it is possible that compositional differences at the species level
164 may account for differences in the size classes recorded.

165 Alternative methods of dietary analysis are recommended in future studies of diving
166 duck diet at Lough Neagh, as oesophageal content analysis of a large number of individuals
167 resulted in a limited sample size. Of those, stable isotope analysis (Inger & Bearhop 2008),
168 which allows for the inclusion of all 79 diving duck individuals irrespective of their
169 oesophageal content, is currently being employed to study the diet of ducks in greater detail.

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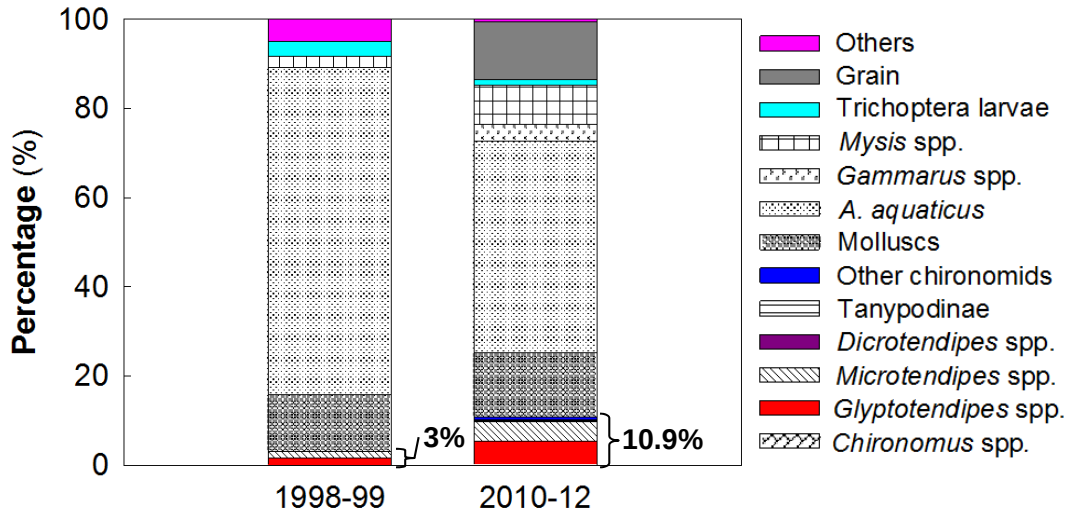

Figure 1. Comparison of Tufted Duck diet between 1998-99 (Bigsby 2000) and 2010-12. The percentages indicate the combined contribution of chironomid larvae.

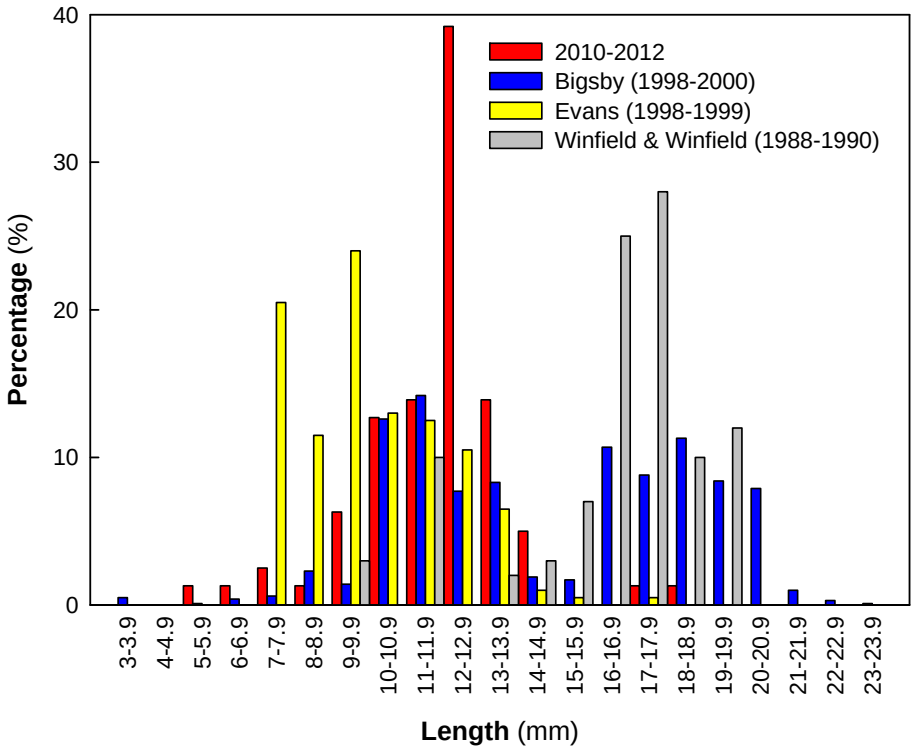

Figure 2. Comparison of length frequencies of chironomid larvae from Tufted Duck diet during 1988-1990, 1998-1999, 1998-2000 (Winfield & Winfield 1994, Evans 2000, Bigsby 2000) and 2010-2012.